



In type 1 diabetes, the body's immune cells mercilessly attack insulin-producing cells like the red cells shown in this image that are clumped together to form a pancreatic islet. University of Pittsburgh scientists have figured out how to disarm the attackers and could soon be the fathers of a cure.

A PROMISING TREATMENT FOR TYPE 1 DIABETES,  
IN THE LAST PLACE ANYONE WOULD EXPECT TO  
FIND IT | BY MELINDA WENNER

# DIABETES TRIALS

It looks a lot like any other hospital room. There's a bed, sink, bench, and a closet overflowing with gloves and syringes. On the wall hangs a poster about diabetes and an advertisement for the University of Pittsburgh Medical Center, which, the poster reminds its readers, is the number one hospital system in the region. But in this room on the sixth floor of UPMC Montefiore, 14 people are changing history.

They're volunteers in a phase I clinical trial that, at first glance, looks like the worst idea ever: In an attempt to calm the overactive immune response that causes type 1 diabetes, doctors are injecting the volunteers with cells notorious for doing the exact opposite. In other words, the patients are being pumped full of something that would normally make them much, much sicker. But no one—not the volunteers, not their families, not the doctors—seems worried. In fact, everyone seems excited: They might just be testing the first diabetes cure.

A disease that plagues more than a million Americans, type 1 diabetes is no picnic. Sufferers must inject themselves with insulin daily just to stay alive, which means that 36-year-old Chad Shumaker, one of the volunteers in the Pitt trial, has given himself nearly 14,000 injections since he was first diagnosed 19 years ago.

“Giving yourself shots twice a day loses its appeal after the first few,” he says.

What’s more, later in life, Shumaker and others with diabetes are at a heightened risk of developing blindness, nerve damage, and heart disease. At its core, the illness (not to be confused with its more common cousin, type 2 diabetes) is a disease of the immune system, which, thanks to a combination of genetic and environmental triggers, mistakenly attacks and kills the cells in the pancreas responsible for producing the hormone insulin. Without insulin, the body cannot store sugar and fat for energy and will eventually fall into a coma.

Shumaker, who owns a garage-door company and has just built a new home in Monroeville, Pa., doesn’t yet know whether he’s being treated with the “real deal” experimental treatment or a placebo in this study that will determine the safety, but not the

**M**assimo Trucco and Nick Giannoukakis could not be more different. Trucco is the Hillman Professor of Pediatric Immunology and an MD professor of pediatrics, pathology, human genetics, and epidemiology at the University of Pittsburgh who heads the Division of Immunogenetics at Children’s Hospital of Pittsburgh of UPMC. He is easily excited, has a guttural laugh that slips out frequently, and speaks with a strong Italian accent occasionally peppered with a “bravissimo!” Giannoukakis, who has a PhD in endocrine genetics and is an associate professor of pathology and immunology at Pitt, was raised in Montreal, speaks crisply and carefully, and emphasizes that science is less about achievements than about making careful choices.

The two do, however, have something important in common: Both like to tackle only the most difficult problems.

“When someone says, ‘Oh, that doesn’t work,’ or, ‘Oh, that’s impossible,’ I say, ‘Okay, we have to do that,’” Trucco explains. Although scientists have been working on a cure for type 1 diabetes for decades, efforts have so far yielded little.

“It’s easy to activate the immune system,”

cells say, “Hey, you know what? You better go back to where I’m coming from because there’s trouble over there.”

That’s when the genetic predisposition to type 1 diabetes comes into play. Normally, after the dendritic police instruct the T cells to attack (these are rough cops), the body performs a quick check to ensure that its T cells have not just been told to pummel something made by the body itself. People born with a genetic predisposition to diabetes, however, have slightly misshapen T cells, so they don’t get the body’s message that the beta cells are innocent, well-meaning guys. Soon enough, the T cells are sent off to work over the beta cells, which surrender and stop producing insulin. The person has developed type 1 diabetes, and the cycle continues indefinitely.

Trucco and Giannoukakis figured there had to be a way to intervene. Perhaps they could manipulate the body’s police in a way that would prevent them from communicating with T cells. T cells become activated in two steps, both involving contact with proteins on the surface of dendritic cells. Without the second interaction, there will be no T-cell response. In fact, the T cells will self-destruct or become quiescent.

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effectiveness, of the new approach. He’s probably too old to get much benefit out of such a treatment anyway. It’s likely that the illness has already killed off too many of his insulin-producing cells to make a full recovery possible. But he’s not participating in the trial for himself, so he doesn’t really mind.

“I’ve got two kids,” he says—Merrick, 1, and Declan, 5. “If the disease passes on, I’d like to see them have a cure.”

Although a cure might seem simple—stop the immune response already!—it has been anything but. Suppress general immunity, and patients suffer deadly infections. Replenish dead pancreatic cells with healthy ones, and the body mercilessly attacks them, too. To solve the problem, scientists must intervene in the attack without affecting other crucial immune functions, a feat many thought impossible. But with a plan to outsmart the very cells that do diabetes’ dirty work, two Pitt doctors may soon prevail. Their approach has already cured mice of the disease. Soon, thanks to Shumaker and other volunteers, they’ll know whether it can cure people, too.

Giannoukakis says. “You put yourself inside a cold or a hot room, and—guess what?—you activate your immune system.” It’s much, much harder to calm it. So the two doctors knew they had to give it a shot.

Type 1 diabetes is not a simple disease. First, an environmental trigger such as a viral infection causes insulin-producing beta cells in the pancreas to become inflamed. It doesn’t take much—the cells are already “overworked factories,” Giannoukakis says, chugging out insulin every time we swallow even a handful of M&Ms. When the beta cells become inflamed, they catch the eye of specialized immune cells called dendritic cells, whose job it is to patrol the body to look for problems, much like a police officer who patrols a neighborhood.

When the dendritic cells notice that all is not right in the pancreas, they assume the beta cells are the culprit—perhaps they are dangerous intruders. The dendritic cells then travel to the lymph nodes to summon backup from other immune cells, including T cells. As Giannoukakis puts it, the dendritic

Trucco and Giannoukakis took advantage of this loophole that nature provides, treating a patient’s dendritic cells in the lab with short DNA sequences called antisense oligonucleotides, designed to down-regulate the levels of the proteins involved in that second interaction with T cells.

The two scientists went to work to devise their concoction. (Trucco thinks research isn’t all that different from cooking: “The difference is, when you’re cooking, you eat the result, while in the other case, you publish it.”)

Here’s what they came up with: Draw blood from one arm of the patient, pass it through a machine that separates out and retains all the progenitors of dendritic cells, and return the rest of the blood via the other arm. Then take the dendritic cell progenitors to a therapeutic lab at the University of Pittsburgh Cancer Institute, where collaborator Theresa Whiteside, Pitt professor of pathology, grows the dendritic cells in the presence of antisense oligonucleotides according to the Trucco/Giannoukakis recipe. Inject these cells back into the patient’s abdomen, near the pancreas, where they replace or

take over the functions of their naturally occurring counterparts inside the pancreas.

The altered dendritic cells start redirecting immune cell traffic—including the T cells—which either leave the pancreas, become inactive, or die.

And because the dendritic cells are injected under the skin in an area that serves as a fast-track highway to the pancreatic precinct's police station, and do not stray far from there, the treatment doesn't affect the policing cells in the rest of the body, which are crucial for fighting infections.

Type 1 diabetes could, therefore, have a cure, if all goes well with the ongoing phase I safety trial in which Shumaker is participating—it began in July 2007—and subsequent trials. What's most ingenious (and, some might add, risky) about the Pitt approach is that it engineers dendritic cells to perform a task they've never been asked to do. Past dendritic cell treatments have always activated, rather than suppressed, the immune system to treat diseases like cancer. Obviously, an activating treatment in type 1 diabetics would be bad news—it would worsen the immune attack rather than alleviate it. But both doctors have faith that their cells will do the right thing.

**A**t the moment, Trucco and Giannoukakis' approach is not ideal. The first step—when the blood is collected for the eventual generation of dendritic cells—requires patients to lie still for almost three hours. They'd like to offer patients something more convenient.

In 2002, Giannoukakis flew to Boston for the annual conference of the American Society of Gene Therapy. One of the talks caught his eye because it had to do with dendritic cells and microspheres—tiny balls, each about the size of a bacterium, that can be used to deliver drugs or engineered genes. The talk focused on using microspheres in cancer treatments, which wasn't exactly pertinent to his work. Still, Giannoukakis popped in.

The first two talks were irrelevant to Giannoukakis' interests. Yawn. But then the third speaker walked up to the podium. The scientist began his talk by comparing different formulations of these microspheres and remarked that one of them was neutral with regard to the immune system. Giannoukakis immediately perked up. Neutral microspheres? So the exterior of the microspheres did not contain chemicals that would startle the dendritic cells into activating. Maybe the little balls could deliver the

antisense oligonucleotides directly into the pancreas; then, the patrolling dendritic cells could ingest the oligonucleotides inside the patient and achieve the same result as the treatment requiring patients to stay put for three hours. Giannoukakis ran back to his hotel room to e-mail the company that made the neutral microspheres, Epic Therapeutics, now a subsidiary of Baxter Healthcare Corporation. Less than 48 hours later, he met the company's chief technology officer, Larry Brown.

Brown, who happens to have type 1 diabetes, was eager to help. Brown and Giannoukakis discussed how to marry Pitt's oligonucleotides with Epic's technology. Then Brown went back to his lab and built what are known as PROMAXX technology microspheres that would deliver the payload Giannoukakis needed. When Giannoukakis added the microspheres to dendritic cells in a petri dish, the cells ate them and digested the payload, rendering them unable to stimulate T cells. Perfect. Then they tried the microspheres (which Trucco affectionately refers to as onions, one of his favorite vegetables) in diabetic mice, performing the experiment five times in a row. Worked every time.

Giannoukakis and Trucco wrote a proposal to have the new vaccine tested by TrialNet, a network of 18 clinical centers around the world that screen only the most promising new treatments for type 1 diabetes.

They reasoned that if the safety trial using the patient's dendritic cells were a success, TrialNet would be more enthusiastic about testing the microspheres head-to-head against the injected dendritic cells.

TrialNet agreed: Assuming the microspheres are shown to be safe and Trucco and Giannoukakis successfully complete one last experiment, TrialNet will sponsor a phase II clinical trial comparing the two vaccine approaches. The scientists are eager to realize this as soon as possible.

Even if these approaches work, potential limitations loom. First, how long will the treatments last? No one knows how long dendritic cells or regulatory T cells live in the human body, but they eventually die. New, immune-attacking populations could then take over again. Patients might have to return to the hospital for boosters once or twice a year. The phase II TrialNet studies will help answer that question.

The treatments also won't be for everyone. They'll only work in patients who still have some remaining healthy beta cells—in

other words, patients who've recently been diagnosed. It's not going to do much good to stop the immune response against beta cells if no beta cells are left to make more insulin. That said, these treatments might be paired with beta cell transplantations or stem cell treatments, which would help restore insulin production.

Trucco and Giannoukakis hope that the treatments will not only cure diabetic patients, but one day will also prevent the disease in those destined to develop diabetes. Children who are born with the genetic disposition and suffer one of the many potential environmental insults that cause the beta cells to become inflamed are sure to end up with the disease. Because there's a way to test for both the genes and the inflammation, Giannoukakis and Trucco envision screening children whose parents or siblings have type 1 diabetes and treating those at risk to prevent the disease from ever developing.

**W**ith a potential cure for such a devastating illness in hand, you'd think Trucco and Giannoukakis would be jumping for joy. Well, Trucco is always high energy—he hardly ever sleeps, preferring instead to wander around his house thinking and occasionally indulging in pots of spaghetti at 3 a.m. (He wakes up his wife each time. "Eating spaghetti alone is bad," he says.) Giannoukakis says that he most enjoys the beginning of an experiment, when the glint of a potential solution flashes in front of him as it did in Boston that day in 2002. And he argues that he and Trucco aren't the people to thank should the treatments prove successful.

"We can sit and we can theorize and we can intellectualize and we can treat a thousand mice, but at the end of the day, the real hero is the human volunteer," he says. "If anything does work out, they are the people who should be thanked first."

Shumaker doesn't think of himself as a hero. "I figured, *What the hell? Somebody needs to do it,*" he says of his decision to volunteer, which requires a total of 24 visits to the hospital. But he's well aware of how momentous the Pitt trial is.

"It's the first good opportunity they've had at a cure for diabetes, so it's something exciting to be involved in," he says, noting that he hasn't experienced any side effects.

"No extra heads or arms or anything growing out of me," he says with a chuckle, "so it's a success all the way around." ■